



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

BOARD OF PATENT APPEALS AND INTERFERENCES

Applicant: Nicolas VOUTE *et al.*

Title: SMALL DENSE MICROPOROUS SOLID SUPPORT MATERIALS,  
THEIR PREPARATION, AND USE FOR PURIFICATION OF LARGE  
MACROMOLECULES AND BIOPARTICLES

Appl. No.: 09/274,014

Filing Date: March 22, 1999

Examiner: D. Sorkin

Art Unit: 1723

**BRIEF ON APPEAL**

11/05/2003 RHARIS1 00000065 09274014  
01 FC:2401 165.00 0P

Adjustment Dates 11/05/2003 RHARIS1  
11/05/2003 RHARIS1 00000065 09274014  
01 FC:2401 -165.00 0P

11/05/2003 RHARIS1 00000065 09274014  
01 FC:2402 165.00 0P



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE  
**BOARD OF PATENT APPEALS AND INTERFERENCES**

Attorney Docket No. 035394/0117

Applicant: Nicolas VOUTE *et al.*  
Title: SMALL DENSE MICROPOROUS SOLID SUPPORT MATERIALS,  
THEIR PREPARATION, AND USE FOR PURIFICATION OF LARGE  
MACROMOLECULES AND BIOPARTICLES  
Appl. No.: 09/274,014  
Filing Date: March 22, 1999  
Examiner: D. Sorkin  
Art Unit: 1723

**APPELLANTS' BRIEF UNDER 37 CFR §1.192**

Commissioner of Patents  
P.O. Box 1450  
Alexandria, VA

Sir:

This brief is in furtherance of the Notice of appeal filed in this case on July 7, 2003. The fees required under 37 CFR §1.17(f) are included in the attached check. Please charge any fee deficiency or credit any overpayment to Deposit Account 19-0741.

This brief is transmitted in triplicate in conformance with 37 CFR §1.192(a).

***I. REAL PARTY IN INTEREST***

The real party in interest in this case is CIPHERGEN Biosystems, Inc., as evidenced by a chain of title submitted on May 9, 2003, and recorded in the USPTO Assignment Division.

## ***II. RELATED APPEALS AND INTERFERENCES***

Appellants, appellants' legal representatives, and the assignee are aware of no appeal or interference which will directly affect or be directly affected by or have a bearing on the Board's decision in this appeal.

## ***III. STATUS OF CLAIMS***

Pending: 1-66

Withdrawn: 23-58 and 64-66

Rejected: 1-22 and 59-63

Appealed: 1-22 and 59-63

## ***IV. STATUS OF AMENDMENTS***

All claim amendments and requests for reconsideration have been entered.

## ***V. BACKGROUND AND SUMMARY OF THE INVENTION***

High molecular weight ("HMW") macromolecules such as nucleic acids, polysaccharides, protein aggregates, and bioparticles such as viruses, viral vectors, membrane proteins and cellular structures, are difficult to isolate from biological sources due to their physical characteristics. Classical techniques for isolating HMW macromolecules and bioparticles include gradient density centrifugation, microfiltration, ultrafiltration and chromatography. These methods present a number of practical disadvantages. Gradient density centrifugation is a time consuming and energy intensive process and provides only limited purification due to intrinsic molecular or bioparticle heterogeneities. Green *et al.*, "Preparative purification of supercoiled plasmid DNA for therapeutic applications," *Biopharm.* 1997: 5262. Membrane technologies, such as cross flow filtration, require a substantial shear stress to maintain permeate flux and these levels of sheer stress are prejudicial to the integrity of the molecules or particles and

consequently to their biological activities. Braas *et al.*, "Strategies for the isolation and purification of retroviral vectors for gene therapy," *Bioseparation* 6: 211-228 (1996).<sup>1</sup>

Packed bed chromatography and adsorption of large molecular weight molecules or particles are also hampered by the physical characteristics of these compounds, setting stringent limitations in terms of operating bed capacity and pressure drop. On the one hand, these large biological structures do not penetrate into classical gel media commonly used in bioseparation and, as a consequence, these large biological structures do not access the internal surface area and pore volume, where the majority of the adsorptive sites are located. Therefore, the partitioning between mobile and liquid phase and the binding capacity is inherently limited. On the other hand, there is no interest in producing media with pores large enough to accommodate these large or HMW biological structures because the intraparticle diffusion in the pores of such media would be extremely limited due to their large size. Consequently the mass transfer and the productivity of such media would be low.<sup>2</sup>

Therefore, chromatography and adsorption of very large molecular weight molecules and bioparticles are hampered by a screening effect, independent of the mode of adsorption. If adsorption of the target HMW compounds occurs, it is restricted only to the external surface area of sorbent beads, and therefore yields low binding capacities. This mode of operation, known as positive adsorption, is rarely used due to this very low binding capacity.

Direct recovery of large macromolecules in the flow-through of solid phase beds is known as negative solid phase purification. HMW compounds flow through the column without being delayed, while smaller contaminants, like proteins, amino acids, sugars and salts, diffuse in the intraparticle volume of the solid phase porous beads, where they can be delayed or adsorbed. This approach shows numerous drawbacks detrimental to performance of separations. First, if separation is based on size exclusion, the loading and the operational linear velocity are very low, dramatically reducing the column productivity. In addition, if separation is based on adsorption, large resin volumes are required as all the

---

<sup>1</sup> Specification at page 1, lines 8-22.

<sup>2</sup> Specification at page 1, line 23 to page 2, line 2.

contaminants must diffuse and be adsorbed into the beads. Furthermore, negative purification processes do not offer any selectivity between different types of very large macromolecules, as they co-elute in the flow-through. In particular, it is impossible to segregate plasmids from genomic DNA and large RNA molecules using negative chromatography purification processes.<sup>3</sup>

As an intermediate case between positive and negative adsorption processes, the operating conditions can be set such that both the HMW compounds and the contaminants are adsorbed. In this situation, flow-through of the target component (such as a very large macromolecule) will occur only after the initial saturation of the external surface of the beads. Such conditions, however, lead to a decrease in target component recovery.<sup>4</sup>

In addition, solutions of HMW biopolymers (such as nucleic acids and polysaccharides) and bioparticles tend to have a high viscosity. In turn, the high viscosity impairs purification of these compounds in many ways. For example, it reduces the diffusivity of the compounds, and therefore tremendously reduces boundary layer and intraparticle mass transfer rate. It also increases the hydraulic resistance of a fixed bed column and generates large pressure drops.<sup>5</sup>

The augmentation of mass transfer resistance is extremely prejudicial to the adsorbent capture efficiency. Longer residence times can potentially counterbalance the reduced rate of adsorption. In order to achieve such longer residence time, however, it would be necessary to use very low linear velocity or very long columns. Both strategies are impracticable as they result in very long purification cycle time and increased pressure drop.<sup>6</sup>

Large pressure drops generated by high viscosity samples, such as those containing HMW macromolecules, restrict the use of semi-rigid adsorbents as these semi-rigid adsorbents are deformed under the mechanical strain and lead to clogging of the column.

---

<sup>3</sup> Specification at page 2, lines 3-21.

<sup>4</sup> Specification at page 2, lines 22-27.

<sup>5</sup> Specification at page 2, line 28 to page 3, line 2.

<sup>6</sup> Specification at page 3, lines 3-8.

In order to reduce the pressure drop, extremely low flow rates or very large particle diameter could be used. However, at the preparative level, both solutions are unrealistic because they lead to large cycle time on the one hand, and very low binding capacity due to too small interactive surface area of large bioparticles on the other hand. Furthermore, solid particles injected through a packed bed of beads are progressively trapped in the intraparticle spaces where they accumulate and tend to irreversibly clog the column.<sup>7</sup>

Some of the problems associated with high viscosity samples and the presence of particulates in a feed stock can be circumvented by using a stirred tank. However, the solid and liquid mixing using stirred tank contactors restrict the capture efficiency. Compared to a fixed bed, the productivity of a stirred tank is reduced due to the low concentration of the adsorbent in the contactor. Moreover, semi-open systems, such as stirred tanks, are difficult to clean, sanitize and automate.<sup>8</sup>

Fluidized bed contactors are also an alternative means for processing high viscosity samples and samples containing insoluble particles. See, *e.g.*, Buijs and Wesselingh, "Batch Fluidized ion-exchange column for stream containing suspended particles," *J. Chrom.*, 201: 319-327 (1980); Chase "Purification of proteins by adsorption chromatography in expanded beds," *Tibtec*, 12: 296-303 (1994); Somers *et al.*, "Isolation and purification of endo-polygalacturonase by affinity chromatography in a fluidized bed reactor," *Chem. Eng. J.* 40: B7-B19 (1989); and Wells *et al.*, "Liquid fluidized bed adsorption in biochemical recovery from biological suspensions," *in* SEPARATION FOR BIOTECHNOLOGY 217-224 (M. Verall, ed.) (1987). However, the media or adsorbents commercially available at present are inadequate for the purification of HMW molecules and particles. See U.S. patent No. 5,522,993 and European patents EP 0 538 350 B1 and EP 0 607 998 B1. The internal porosity of these media or adsorbents is inaccessible for very large solutes, and their large particle diameter undesirably decreases the external surface area. As a result, these media provide only limited capacity for the purification of HMW molecules and particles.<sup>9</sup>

---

<sup>7</sup> Specification at page 3, lines 9-19.

<sup>8</sup> Specification at page 3, lines 20-25.

<sup>9</sup> Specification at page 3, line 26 to page 4, line 8.

Fluid bed separation processes are attractive for the recovery of bioproducts as they achieve lower operational pressures than a packed bed and are resistant to fouling by particulates and suspended materials in the feed stock. Fluidized-bed technology has been successfully employed as early as 1958 for the recovery of small molecules, such as antibiotics. (See Bartels *et al.*, "A novel ion exchange method for the isolation of streptomycin," *Chem. Eng. Prog.*, 54(8):49-51 (1958); Belter *et al.*, "Development of a recovery process for novobiocin," *Biotechnol. Bioeng.*, 15:533-549 (1973).) More recently, this technology has been applied for the recovery of larger molecular weight molecules, such as proteins, from unclarified feed stocks. See, A. Bascoul, "Fluidisation liquide-solide. Etude hydrodynamique et extraction des proteines," These d'etat, Universite Paul Sabatier, Toulouse, France (1989); B. Biscans, "Chromatographie d'echange d'ions en couche fluidisee. Extraction des proteines du lactoserum," These de docteur ingénieur, Institut national polytechnique de Toulouse, Toulouse, France (1985); Biscans *et al.*, *Entropie*, 125/126: 27-34 (1985); Biscans *et al.*, *Entropie*, 125/126: 17-26 (1985); Draeger and Chase, "Liquid fluidized bed adsorption of protein in the presence of cells," *Bioseparation*, 2: 67-80 (1991); Draeger and Chase, "Liquid fluidized beds for protein purification," *Trans IChemE*, 69(part C): 45-53 (1991); J. van der Weil, "Continuous recovery of bioproducts by adsorption," PhD Thesis, Delft University, Delft (1989); and Wells *et al.* (1987), *supra*.<sup>10</sup>

United States patent No. 4,976,865 describes a method and a column for fluidized bed chromatographic separation of samples containing molecules which have a tendency towards autodenaturation, including biopolymers of medium molecular weight, such as proteins, enzymes, toxins and antibodies. This method assumes that any suspended material in the sample or feed stock is removed during loading and washing, while the molecules of interest diffuse inside the adsorbent loaded in the column. Yet the operational binding capacity of the procedure and materials describe in U.S. patent No. 4,976,865 are inadequate for the biopurification of HMW molecules and bioparticles.<sup>11</sup>

---

<sup>10</sup> Specification at page 4, lines 9-30.

<sup>11</sup> Specification at page 4, line 31 to page 5, line 6.

United States patent No. 5,522,993 and European patents EP 0 538 350 B1 and EP 0 607 998 B1 describe special polymeric resin media, especially agarose, having small particles of dense materials within the media, and their use in fluidized beds. The dense material described for use trapped within the polymeric resin media include glass, quartz and silica. Despite the gain in density of this media, however, due to the presence of the small particles of dense material, the density is still relatively low; hence, in order to achieve a stabilized fluidized bed, large bead diameter is required to compensate for the low density differential between the liquid and solid phases. The aforementioned European patents also describe beads which consist of a porous conglomerate of polymeric material and density controlling particles therein. The beads described in these three patents are inadequate for the isolation of HMW molecules and bioparticles as the low density and the large particle size of these beads are not conducive to separation of HMW macromolecules and bioparticles.<sup>12</sup>

The present invention provides new dense mineral oxide solid supports or microbeads which exhibit high density, low porosity, high external surface area and high binding capacity. The small dense mineral oxide solid supports or microbeads of the present invention may be used in various solid phase adsorption and chromatography methods including packed bed and fluidized bed methods, and are particularly useful in fluidized bed devices and allow higher linear velocities to be used in such fluidized bed devices. These solid supports or microbeads are particularly suited for separating or isolating large biological molecules, such as bioparticles and high molecule weight macromolecules, especially in fluidized bed or expanded bed methods.<sup>13</sup>

The dense mineral oxide solid supports according to the invention comprise:

(a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and

---

<sup>12</sup> Specification at page 5, lines 7-19.

<sup>13</sup> Specification at page 5, lines 21-30.



(b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support.<sup>14</sup>

In a preferred embodiment, the dense mineral oxide solid supports have a density of 2.1 to 11, and a particle size of 10  $\mu\text{m}$  to 100  $\mu\text{m}$ .<sup>15</sup> The mineral oxide matrix may comprise, *inter alia*, titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof. The interactive polymer network may comprise, *inter alia*, a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.<sup>16</sup>

## ***VI. ISSUES***

The single issue on appeal is whether claims 1-22 and 59-63 would have been obvious over Girot *et al.* (U.S. 5,445,732) alone or combined with Davis, Jr. *et al.* (U.S. 4,203,772).

## ***VII. GROUPS OF CLAIMS***

For purposes of the present appeal, the claims do not all stand or fall together. The following groups of claims will be argued separately:

- |         |   |
|---------|---|
| Group 1 | Claims 1, 9, 10, and 13-22  |
| Group 2 | Claims 11 and 12 (a subset of the claims that recite polysaccharide polymer in the pores) |
| Group 3 | Claims 7, 8 and 60 (a subset of the claims that recite a specified range for pore volume) |
| Group 4 | Claims 2, 3, 6 and 59-63 (a subset of the claims that recite a density for the particles) |

---

<sup>14</sup> See claim 1.

<sup>15</sup> See claim 6.

<sup>16</sup> See claims 9 and 10, respectively.

Thus, the arguments in Section IX.A of the brief apply equally to all of the claims on appeal. The arguments in Sections IX.C.1, IX.C.2, and IX.C.3 of the brief apply to the features that are recited more particularly in Claim Groups 2, 3, and 4, respectively. Accordingly, the arguments advanced in the latter sections additionally support the patentability of claims in Groups 2, 3, and 4, respectively. Even if the arguments that pertain to all appealed claims are not deemed persuasive, the Board is obliged to consider separately the patentability of the claims in each of Groups 2, 3, and 4, in light of the additional arguments presented.

### ***VIII. SUMMARY OF THE ARGUMENT***

Giroto *et al.* (U.S. 5,445,732) would not have suggested a dense mineral oxide solid supports comprising a mineral oxide matrix a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support. Both the patent itself and a declaration from one its inventors clarify that the purpose of invention described in Giroto ('732) was to make the pore volume as large as possible so that molecules would diffuse inside the pores of the matrix. This is consistent with calculated pore volumes of 40 to 60% for Giroto ('732). This approach contrasts sharply with that described in the application, which strives to foreclose access to the pore volume by the molecules in the source solution, using a small pore volume so that a crosslinked polymer can be stably rooted in the matrix and so that interactions should occur on the surface of the bead.

The addition of Davis, Jr. to Giroto ('732) does not gainsay this conclusion. While Davis, Jr. discloses mineral oxide matrices having porosities less than 30%, it would not have been obvious to use the matrix material of Davis, Jr. in place of the matrix disclosed in Giroto. Such a substitution would be contrary to Giroto's purpose of providing beads in which biomolecules penetrate "an open, flexible. . . three-dimensional polymer network" that is contained in the pores. If a proposed modification renders a prior art invention unsatisfactory for its intended purpose, then there is no suggestion or motivation to make

the proposed modification. MPEP §2143.01 and *In re Gordon*, 221 USPQ 1125 (Fed. Cir. 1984).

## **IX. ARGUMENT**

- A. Claims 1-22 and 59-63 would not have been obvious based on Girot *et al.* (U.S. 5,445,732)

Claims 1-22 and 59-63 are rejected under Section 103(a) based on Girot *et al.* The examiner states that:

Girot ('732) discloses dense mineral oxide supports comprising a mineral oxide matrix having an external surface and pores; and an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix. Girot ('732) does not explicitly disclose the porosity being 'less than 30% of the total volume of the mineral oxide matrix' as claimed. However, Girot ('732) recognizes that a broad range of porosities are suitable for the invention [citing col. 8, lines 40-45; col. 15, line 45; and col. 36, lines 12-15]

A dense mineral oxide solid support comprising a mineral oxide matrix having an external surface and pores, in which the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix would not have been suggested by Girot ('732). Girot ('732) discloses, in column 8, that:

Typically, the present invention utilizes base matrices having the following characteristics: an initial average particle size ranging from about 5 to about 1000 microns; an initial porous volume ranging from about 0.2 to about 2 cm<sup>3</sup>/gram; an initial surface area ranging from about 1 to about 800 m<sup>2</sup>/gram; and an initial pore size ranging from about 50 to about 6000 angstroms. Preferably, the base matrix is characterized by: an initial average particle size ranging from about 10 to about 300 microns, although passivated supports having narrow particle size ranges, such as about 15-20, about 15-25, about 30-45, about 50-60, about 80-100, and about 100-300 microns, are most preferred. Preferred ranges for other characteristics include an initial porous volume ranging from about 0.8 to about 1.2 cm<sup>3</sup>/gram; an initial surface area ranging from about 10 to about 400

m<sup>2</sup>/gram; and an initial pore size ranging from about 1000 to about 3000 angstroms. The density of the porous solid matrix obviously varies with its chemical nature, being higher for mineral oxide (*e.g.*, silica) substrates and lower for polymeric ones (*e.g.*, polystyrene).

The use of pore volumes ranging from 0.8 to 1.2 cm<sup>3</sup>/gr of solid material marks a fundamental difference between Girot ('732) and the present invention. Girot ('732) relates primarily to porous silica, for which a porous volume of 0.8 to 1.2 cm<sup>3</sup>/gr equates to a pore volume of between 0.4 to 0.6 ml per ml of solid material or between 40 and 60% (porous silica has a volume/weight ratio of 2, *i.e.*, 2 ml or 2 cm<sup>3</sup> of beads yield about 1 gram of material).

This is consistent with attestations in a declaration of Dr. Boschetti that was submitted during prosecution before the examiner. Dr. Boschetti is a co-inventor of Girot ('732) and thus eminently qualified to inform as to its content. Dr. Boschetti attests that the purpose of invention described in Girot ('732) was to have a hydrogel fill the pores entirely, while allowing even macromolecules still to diffuse inside. Therefore, the pore volume is made as large as possible, to maximize the binding capacity of the final material. The pore volume of 40 to 60% calculated above is in line with this purpose. Viewed in its proper context, which takes into account the purposed of the invention in Girot ('732), the disclosure of Girot ('732) would not have led one of ordinary skill to decrease porosity, or pore volume, below 30%. This approach contrasts sharply with that described in the application. The latter strives to foreclose access to the pore volume by the molecules in the source solution, and it uses a small pore volume in order that a crosslinked polymer can be stably rooted in the matrix and that interactions should occur on the surface of the bead.

This also is supported by an explanation contained in Girot ('732) that relate to the underlying mechanism of the invention. Thus, at column 19 it is disclosed that:

It is believed, without wishing to be limited by theory, that a highly open, flexible lattice structure comprised primarily of polymeric chains of repeating main monomer units is formed within the pores of the porous solid matrix. Very significantly, it is believed that the areas of the porous support available for desirable reversible interaction with

biological molecules are not confined to the regions immediately adjacent to the surface of the pore as is the case when thin, substantially two-dimensional coatings are applied to porous surfaces in the manner of Steuck (U.S. Pat. No. 4,618,533) and Varady *et al.* (U.S. Pat. No. 5,030,352) as discussed in Section 2.2 above. Rather, it is believed that the polymeric network of the present invention extends outwardly into the pore volume itself in the manner of a three-dimensional lattice, as opposed to a two-dimensional coating limited strictly to the pore wall surface area. A schematic diagram of such a structure, as it is thought to exist, is illustrated in FIG. 5, where a biological molecule of interest (depicted as a spherical object) is also shown interacting with the lattice. Furthermore, the presence of porogens (pore-inducers) in the passivation mixture is believed to promote creation of this open three-dimensional polymer network.

It is thought that perhaps the open, flexible nature of the three-dimensional polymer network allows biological molecules *to rapidly penetrate the polymer lattice and thereby efficiently interact with sorptive groups in the polymer network of the passivated porous support* even at high solution flowrates. The *rapid and efficient mass transfer of biomolecules into and through this network* avoids the decrease in useful or dynamic sorption capacity and resolution that are typical of conventional chromatographic media. With these conventional media, diffusion in the pores of the support and/or materials coated thereupon or within them leads to poor mass transfer rates and limits the efficiency of the chromatographic process.

(Emphasis added.)

This approach, in which biomolecules enter the pores of a mineral oxide bead, contrasts sharply with the present invention, in which pore volume is minimized in order to foreclose access to the pore volume by molecules in the solution. The low porosity as presently claimed allows a crosslinked polymer to be stably rooted in the matrix, "so that subsequent interaction with macromolecules occurs on the external surface area of the support." This is clearly contrary to both the purpose of Girot ('732) and to the physical structure taught by Girot ('732).

The examiner argues that the skilled artisan would have been motivated to minimize porosity in light of Girot's teaching that "it is generally desirable to have as great

a density difference as possible between the solid support particles and the fluidizing medium.” This teaching in Girot (‘732) must be balanced, however, against the primary purpose of Girot (‘732), which was to provide a bead in which even macromolecules can enter the pores. When designing beads for fluidized bed chromatography, the skilled artisan might have been motivated to select values at the lower end of the stated pore volume range in Girot (‘732), in order to maximize the density difference. But the person of ordinary skill would *not* have been motivated to extend the range lower than the endpoint of the disclosed range. Such an extension, beyond the range disclosed in Girot (‘732), would directly contravene the ability to have biomolecules diffuse into the pores. A porosity of less than 30% as presently claimed is therefore not a “workable” range when considered in light of the purpose of Girot (‘732), and one of ordinary skill in the art would not have been motivated to use a pore volume less than the endpoint of Girot’s range.

In an Advisory Action dated July 2, 2003, the examiner states that “the instant claims in no way exclude the possibility of macromolecules entering pores.” The instant claims do recite, however, “an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, *so that subsequent interaction with macromolecules occurs on the external surface area of the support*” – claims 1 and 6, emphasis added. This is in distinct contrast to the mechanism proposed in Girot (‘732), in which molecules diffuse into a polymeric lattice contained within the pores, as quoted above. Subsequent interaction on the external surface area of the support is not suggested in Girot (‘732).

The examiner comments in the Advisory Action that “clearly the size of a given pore at a surface entrance (if there is one) determines if a molecule can enter, not the percent porosity. . . a hollow sphere could have >99% porosity and not let anything in.” Certainly, one can invent hypothetical situations to demonstrate that porosity is not equivalent to openness. However, in the real world of mineral oxide beads a relationship between porosity and the ability of molecules to penetrate is recognized. For example, Girot (‘732) notes that “the present invention relates to the passivation of non-passivated porous solid matrices while *maximizing the openness (e.g., gel porosity and pore size)* of the resulting passivated porous support” (column 9, lines 7-8; emphasis added). The

hypothetical situations proposed in the Advisory Action would not be the basis on which a skilled artisan would interpret the teachings of the prior art.

Finally, the examiner's comments in the Advisory Action that "many claims of Girot ('732) are not limited to any particular porosity" and that "claim 13 of Girot ('732) explicitly recites *excluding* molecules as small as 500 daltons" (emphasis in original) do not belie the clear teaching that interaction in Girot ('732) occurs within the polymeric lattice contained within the pores. The fact that molecules of a certain size are excluded in Girot's claims in no way implies that the molecules of interest, *i.e.*, those not excluded, are interacting on the external surface area of the support as presently claimed. Indeed, the very concept of size exclusion supports the conclusion that Girot's mechanism is one in which molecules diffuse into the beads, as clearly proposed by Girot. This understanding of Girot's mechanism is supported by attestations of one of the inventors of Girot ('732). In light of this, the interpretation by the examiner of Girot's teaching clearly is unsupportable.

B. The addition of Davis, Jr. (U.S. 4,203,772) to Girot ('732) would not have suggested the invention as presently recited in claims 1-22 and 59-63

In the alternative, claims 1-22 and 59-63 stand rejected under Section 103(a) based on Girot ('732) in view of Davis *et al.* (U.S. 4,203,772). The examiner relies on Davis, Jr. *et al.* as explicitly disclosing mineral oxide matrices having porosities less than 30%, and argues that it would have been obvious to use the matrix material of Davis, Jr. *et al.* in place of the matrix disclosed in Girot. Once again, this would be contrary to Girot's purpose of providing beads in which biomolecules penetrate "an open, flexible. . . three-dimensional polymer network" that is contained in the pores. As noted in detail above, interaction in Girot is not on the surface as presently claimed. To modify Girot by using the mineral oxide matrix of Davis would be to render Girot unsuitable for its intended purpose. If a proposed modification renders a prior art invention unsatisfactory for its intended purpose, then perforce there would have been no suggestion or motivation to make the proposed modification. MPEP §2143.01 and *In re Gordon*, 221 USPQ 1125 (Fed. Cir. 1984). Accordingly, the modification proposed by the addition of Davis, Jr. to Girot is improper and cannot be sustained.

C. While no *prima facie* case of obviousness exists for any of the claims, additional bases for patentability exist for subgroups of the appealed claims

1. Claims 11 and 12 recite the pores contain a polysaccharide, a feature not suggested by the art of record

Claim 11 recites the soluble organic polymer is a polysaccharide or a mixture of polysaccharides, while claim 12 more particularly recites that the polysaccharide is selected from the group consisting of agarose, dextran, cellulose, chitosan, a glucosaminoglycan, and derivatives thereof. These polymers are quite different than those in Girot ('732), and would not have been suggested by the cited art of record.

2. Claims 7, 8 and 60 recite a ranges for pore volume that would not have been suggested by the art of record

Claims 7 and 60 recite a pore volume is 5% to 25% of the total volume of the mineral oxide matrix, while claim 8 recites a pore volume of 5% to 15%. These smaller pore volumes deviate even further from Girot's teaching of a maximized pore volume than does the recitation in the other claims of pore volumes less than 30%. Accordingly, the case in favor of patentability is even stronger as to these claims.

3. Claims 2, 3, 6 and 60-63 recite a density for the particles that would not have been suggested by the art of record

Claim 2 recites that the dense mineral oxide solid supports have a density in the range of about 1.7 to 11, claim 3 recites a density in the range of about 2.1 to about 10, and claims 6 and 60-63 recite a density of 2.1 to 11. Girot ('732) merely indicates that "density of the porous solid matrix obviously varies with its chemical nature" (column 8, lines 59-60), and provides no guidance on the issue of density. Accordingly, there clearly is no *prima facie* case of obviousness with respect to claims 2, 3, 6 and 60-63.

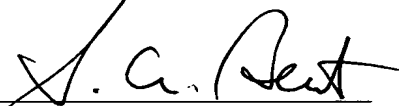


**X. CONCLUSION**

For these reasons, the Board is respectfully requested to reverse the examiner and remand this application for issuance.

Respectfully submitted,

4 November 2002  
Date

  
Stephen A. Bent  
Attorney for Applicant  
Reg. No. 29,768

**FOLEY & LARDNER**  
3000 K Street, N.W., Suite 500  
Washington, D.C. 20007-5109  
Telephone: (202) 672-5492  
Facsimile: (202) 672-5399

**APPENDIX: APPEALED CLAIMS**

What is claimed is:

1. (Twice Amended) Dense mineral oxide solid supports comprising:
  - (a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and
  - (b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support.
2. The dense mineral oxide solid supports of Claim 1, having a density in the range of about 1.7 to 11.
3. The dense mineral oxide solid supports of Claim 2, wherein the density is in the range of about 2.1 to about 10.
4. The dense mineral oxide solid supports of Claim 1, wherein said dense mineral oxide solid supports have a particle size in the range of about 5  $\mu\text{m}$  to about 500  $\mu\text{m}$ .
5. The dense mineral oxide solid supports of Claim 4, wherein the particle size is in the range of about 10  $\mu\text{m}$  to about 100  $\mu\text{m}$ .
6. (Twice Amended) Dense mineral oxide solid supports comprising:
  - (a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and
  - (b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support,

wherein said dense mineral oxide solid supports have a density of 2.1 to 11, and a particle size of 10  $\mu\text{m}$  to 100  $\mu\text{m}$ .

7. The dense mineral oxide solid supports of claim 1, wherein the pore volume is 5% to 25% of the total volume of the mineral oxide matrix.

8. The dense mineral oxide solid supports of Claim 7, wherein the pore volume is 5% to 15%.

9. The dense mineral oxide solid supports of claim 1, wherein the mineral oxide matrix is comprised of titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof.

10. The dense mineral oxide solid supports of claim 1, wherein the interactive polymer network comprises a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.

11. The dense mineral oxide solid supports of Claim 10, wherein the soluble organic polymer is a polysaccharide or a mixture of polysaccharides.

12. The dense mineral oxide solid supports of Claim 11, wherein the polysaccharide is selected from the group consisting of agarose, dextran, cellulose, chitosan, a glucosaminoglycan, and derivatives thereof.

13. The dense mineral oxide solid supports of Claim 10, wherein the soluble organic polymer is a linear soluble organic polymer selected from the group consisting of polyvinyl alcohol, a polyethyleneimine, a polyvinylamine, polyvinylpyrrolidone, a polyethyleneglycol, a polyaminoacid, a polynucleic acid, and derivatives thereof.

14. The dense mineral oxide solid supports of claim 1, wherein the interactive polymer network comprises monomers, bifunctional monomers, or mixtures thereof copolymerized in place with the mineral oxide matrix.

15. The dense mineral oxide solid supports of Claim 14, wherein the monomers are selected from the group consisting of:

(a) aliphatic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds;

(b) aromatic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds;

(c) heterocyclic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds; and

(d) mixtures of any of the monomers in (a), (b) or (c).

16. The dense mineral oxide solid supports of Claim 15, wherein (a) is acrylamide, dimethylacrylamide, trisacryl, acrylic acid, acryloylglycine, diethylaminoethyl methacrylamide, vinylpyrrolidone, vinylsulfonic acid, allylamine, allylglycidylether, or derivatives thereof.

17. The dense mineral oxide solid supports of Claim 15, wherein (b) is vinyltoluene, phenylpropylacrylamide, trimethylaminophenylbutylmethacrylate, tritylacrylamid; or derivatives thereof.

18. The dense mineral oxide solid supports of Claim 15, wherein (c) is vinylimidazole, vinylpyrrolidone, acryloylmorpholine, or derivatives thereof.

19. The dense mineral oxide solid supports of Claim 14, wherein the bifunctional monomers are selected from the group consisting of:

(a) bisacrylamides;

(b) bis-methacrylamides;

(c) bis-acrylates;

(d) ethyleneglycol-methacrylates; and

(e) diallyltartradiamide.

20. The dense mineral oxide solid supports of Claim 19, wherein (a) is N,N'-methylene-bis-acrylamide, N,N'-ethylene-bis-acrylamide, N,N'-hexamethylene-bis-acrylamide, or glyoxal-bis-acrylamide.

21. The dense mineral oxide solid supports of Claim 19, wherein (b) is N,N'-methylene-bis-methacrylamide, N,N'-ethylene-bis-methacrylamide, or N,N'-hexamethylene-bis-methacrylamide.

22. The dense mineral oxide solid supports of Claim 19, wherein (c) is ethyleneglycoldiacrylate, or ethyleneglycoldimethacrylate.

59. The dense mineral oxide solid supports of claim 2, wherein said dense mineral oxide solid supports have a particle size in the range of about 5  $\mu\text{m}$  to about 500  $\mu\text{m}$ .

60. The dense mineral oxide solid supports of claim 6, wherein the pore volume is 5% to 25% of the total volume of the mineral oxide matrix.

61. The dense mineral oxide solid supports of claim 6, wherein the mineral oxide matrix is comprised of titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof.

62. The dense mineral oxide solid supports of claim 6, wherein the interactive polymer network comprises a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.

63. The dense mineral oxide solid supports of claim 6, wherein the interactive polymer network comprises monomers, bifunctional monomers, or mixtures thereof copolymerized in place with the mineral oxide matrix.



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

**BOARD OF PATENT APPEALS AND INTERFERENCES**

Attorney Docket No. 035394/0117

Applicant: Nicolas VOUTE *et al.*

Title: SMALL DENSE MICROPOROUS SOLID SUPPORT MATERIALS,  
THEIR PREPARATION, AND USE FOR PURIFICATION OF LARGE  
MACROMOLECULES AND BIOPARTICLES

Appl. No.: 09/274,014

Filing Date: March 22, 1999

Examiner: D. Sorkin

Art Unit: 1723

**APPELLANTS' BRIEF UNDER 37 CFR §1.192**

Commissioner of Patents  
P.O. Box 1450  
Alexandria, VA

Sir:

This brief is in furtherance of the Notice of appeal filed in this case on July 7, 2003. The fees required under 37 CFR §1.17(f) are included in the attached check. Please charge any fee deficiency or credit any overpayment to Deposit Account 19-0741.

This brief is transmitted in triplicate in conformance with 37 CFR §1.192(a).

***I. REAL PARTY IN INTEREST***

The real party in interest in this case is CIPHERGEN Biosystems, Inc., as evidenced by a chain of title submitted on May 9, 2003, and recorded in the USPTO Assignment Division.

## ***II. RELATED APPEALS AND INTERFERENCES***

Appellants, appellants' legal representatives, and the assignee are aware of no appeal or interference which will directly affect or be directly affected by or have a bearing on the Board's decision in this appeal.

## ***III. STATUS OF CLAIMS***

Pending: 1-66

Withdrawn: 23-58 and 64-66

Rejected: 1-22 and 59-63

Appealed: 1-22 and 59-63

## ***IV. STATUS OF AMENDMENTS***

All claim amendments and requests for reconsideration have been entered.

## ***V. BACKGROUND AND SUMMARY OF THE INVENTION***

High molecular weight ("HMW") macromolecules such as nucleic acids, polysaccharides, protein aggregates, and bioparticles such as viruses, viral vectors, membrane proteins and cellular structures, are difficult to isolate from biological sources due to their physical characteristics. Classical techniques for isolating HMW macromolecules and bioparticles include gradient density centrifugation, microfiltration, ultrafiltration and chromatography. These methods present a number of practical disadvantages. Gradient density centrifugation is a time consuming and energy intensive process and provides only limited purification due to intrinsic molecular or bioparticle heterogeneities. Green *et al.*, "Preparative purification of supercoiled plasmid DNA for therapeutic applications," *Biopharm.* 1997: 5262. Membrane technologies, such as cross flow filtration, require a substantial shear stress to maintain permeate flux and these levels of sheer stress are prejudicial to the integrity of the molecules or particles and

consequently to their biological activities. Braas *et al.*, "Strategies for the isolation and purification of retroviral vectors for gene therapy," *Bioseparation* 6: 211-228 (1996).<sup>1</sup>

Packed bed chromatography and adsorption of large molecular weight molecules or particles are also hampered by the physical characteristics of these compounds, setting stringent limitations in terms of operating bed capacity and pressure drop. On the one hand, these large biological structures do not penetrate into classical gel media commonly used in bioseparation and, as a consequence, these large biological structures do not access the internal surface area and pore volume, where the majority of the adsorptive sites are located. Therefore, the partitioning between mobile and liquid phase and the binding capacity is inherently limited. On the other hand, there is no interest in producing media with pores large enough to accommodate these large or HMW biological structures because the intraparticle diffusion in the pores of such media would be extremely limited due to their large size. Consequently the mass transfer and the productivity of such media would be low.<sup>2</sup>

Therefore, chromatography and adsorption of very large molecular weight molecules and bioparticles are hampered by a screening effect, independent of the mode of adsorption. If adsorption of the target HMW compounds occurs, it is restricted only to the external surface area of sorbent beads, and therefore yields low binding capacities. This mode of operation, known as positive adsorption, is rarely used due to this very low binding capacity.

Direct recovery of large macromolecules in the flow-through of solid phase beds is known as negative solid phase purification. HMW compounds flow through the column without being delayed, while smaller contaminants, like proteins, amino acids, sugars and salts, diffuse in the intraparticle volume of the solid phase porous beads, where they can be delayed or adsorbed. This approach shows numerous drawbacks detrimental to performance of separations. First, if separation is based on size exclusion, the loading and the operational linear velocity are very low, dramatically reducing the column productivity. In addition, if separation is based on adsorption, large resin volumes are required as all the

---

<sup>1</sup> Specification at page 1, lines 8-22.

<sup>2</sup> Specification at page 1, line 23 to page 2, line 2.



contaminants must diffuse and be adsorbed into the beads. Furthermore, negative purification processes do not offer any selectivity between different types of very large macromolecules, as they co-elute in the flow-through. In particular, it is impossible to segregate plasmids from genomic DNA and large RNA molecules using negative chromatography purification processes.<sup>3</sup>

As an intermediate case between positive and negative adsorption processes, the operating conditions can be set such that both the HMW compounds and the contaminants are adsorbed. In this situation, flow-through of the target component (such as a very large macromolecule) will occur only after the initial saturation of the external surface of the beads. Such conditions, however, lead to a decrease in target component recovery.<sup>4</sup>

In addition, solutions of HMW biopolymers (such as nucleic acids and polysaccharides) and bioparticles tend to have a high viscosity. In turn, the high viscosity impairs purification of these compounds in many ways. For example, it reduces the diffusivity of the compounds, and therefore tremendously reduces boundary layer and intraparticle mass transfer rate. It also increases the hydraulic resistance of a fixed bed column and generates large pressure drops.<sup>5</sup>

The augmentation of mass transfer resistance is extremely prejudicial to the adsorbent capture efficiency. Longer residence times can potentially counterbalance the reduced rate of adsorption. In order to achieve such longer residence time, however, it would be necessary to use very low linear velocity or very long columns. Both strategies are impracticable as they result in very long purification cycle time and increased pressure drop.<sup>6</sup>

Large pressure drops generated by high viscosity samples, such as those containing HMW macromolecules, restrict the use of semi-rigid adsorbents as these semi-rigid adsorbents are deformed under the mechanical strain and lead to clogging of the column.

---

<sup>3</sup> Specification at page 2, lines 3-21.

<sup>4</sup> Specification at page 2, lines 22-27.

<sup>5</sup> Specification at page 2, line 28 to page 3, line 2.

<sup>6</sup> Specification at page 3, lines 3-8.

In order to reduce the pressure drop, extremely low flow rates or very large particle diameter could be used. However, at the preparative level, both solutions are unrealistic because they lead to large cycle time on the one hand, and very low binding capacity due to too small interactive surface area of large bioparticles on the other hand. Furthermore, solid particles injected through a packed bed of beads are progressively trapped in the intraparticle spaces where they accumulate and tend to irreversibly clog the column.<sup>7</sup>

Some of the problems associated with high viscosity samples and the presence of particulates in a feed stock can be circumvented by using a stirred tank. However, the solid and liquid mixing using stirred tank contactors restrict the capture efficiency. Compared to a fixed bed, the productivity of a stirred tank is reduced due to the low concentration of the adsorbent in the contactor. Moreover, semi-open systems, such as stirred tanks, are difficult to clean, sanitize and automate.<sup>8</sup>

Fluidized bed contactors are also an alternative means for processing high viscosity samples and samples containing insoluble particles. See, *e.g.*, Buijs and Wesselingh, "Batch Fluidized ion-exchange column for stream containing suspended particles," *J. Chrom.*, 201: 319-327 (1980); Chase "Purification of proteins by adsorption chromatography in expanded beds," *Tibtec*, 12: 296-303 (1994); Somers *et al.*, "Isolation and purification of endo-polygalacturonase by affinity chromatography in a fluidized bed reactor," *Chem. Eng. J.* 40: B7-B19 (1989); and Wells *et al.*, "Liquid fluidized bed adsorption in biochemical recovery from biological suspensions," *in* SEPARATION FOR BIOTECHNOLOGY 217-224 (M. Verall, ed.) (1987). However, the media or adsorbents commercially available at present are inadequate for the purification of HMW molecules and particles. See U.S. patent No. 5,522,993 and European patents EP 0 538 350 B1 and EP 0 607 998 B1. The internal porosity of these media or adsorbents is inaccessible for very large solutes, and their large particle diameter undesirably decreases the external surface area. As a result, these media provide only limited capacity for the purification of HMW molecules and particles.<sup>9</sup>

---

<sup>7</sup> Specification at page 3, lines 9-19.

<sup>8</sup> Specification at page 3, lines 20-25.

<sup>9</sup> Specification at page 3, line 26 to page 4, line 8.

Fluid bed separation processes are attractive for the recovery of bioproducts as they achieve lower operational pressures than a packed bed and are resistant to fouling by particulates and suspended materials in the feed stock. Fluidized-bed technology has been successfully employed as early as 1958 for the recovery of small molecules, such as antibiotics. (See Bartels *et al.*, "A novel ion exchange method for the isolation of streptomycin," *Chem. Eng. Prog.*, 54(8):49-51 (1958); Belter *et al.*, "Development of a recovery process for novobiocin," *Biotechnol. Bioeng.*, 15:533-549 (1973).) More recently, this technology has been applied for the recovery of larger molecular weight molecules, such as proteins, from unclarified feed stocks. See, A. Bascoul, "Fluidisation liquide-solide. Etude hydrodynamique et extraction des proteines," These d'etat, Universite Paul Sabatier, Toulouse, France (1989); B. Biscans, "Chromatographie d'echange d'ions en couche fluidisee. Extraction des proteines du lactoserum," These de docteur ingenieur, Institut national polytechnique de Toulouse, Toulouse, France (1985); Biscans *et al.*, *Entropie*, 125/126: 27-34 (1985); Biscans *et al.*, *Entropie*, 125/126: 17-26 (1985); Draeger and Chase, "Liquid fluidized bed adsorption of protein in the presence of cells," *Bioseparation*, 2: 67-80 (1991); Draeger and Chase, "Liquid fluidized beds for protein purification," *Trans IChemE*, 69(part C): 45-53 (1991); J. van der Weil, "Continuous recovery of bioproducts by adsorption," PhD Thesis, Delft University, Delft (1989); and Wells *et al.* (1987), *supra*.<sup>10</sup>

United States patent No. 4,976,865 describes a method and a column for fluidized bed chromatographic separation of samples containing molecules which have a tendency towards autodenaturation, including biopolymers of medium molecular weight, such as proteins, enzymes, toxins and antibodies. This method assumes that any suspended material in the sample or feed stock is removed during loading and washing, while the molecules of interest diffuse inside the adsorbent loaded in the column. Yet the operational binding capacity of the procedure and materials describe in U.S. patent No. 4,976,865 are inadequate for the biopurification of HMW molecules and bioparticles.<sup>11</sup>

---

<sup>10</sup> Specification at page 4, lines 9-30.

<sup>11</sup> Specification at page 4, line 31 to page 5, line 6.

United States patent No. 5,522,993 and European patents EP 0 538 350 B1 and EP 0 607 998 B1 describe special polymeric resin media, especially agarose, having small particles of dense materials within the media, and their use in fluidized beds. The dense material described for use trapped within the polymeric resin media include glass, quartz and silica. Despite the gain in density of this media, however, due to the presence of the small particles of dense material, the density is still relatively low; hence, in order to achieve a stabilized fluidized bed, large bead diameter is required to compensate for the low density differential between the liquid and solid phases. The aforementioned European patents also describe beads which consist of a porous conglomerate of polymeric material and density controlling particles therein. The beads described in these three patents are inadequate for the isolation of HMW molecules and bioparticles as the low density and the large particle size of these beads are not conducive to separation of HMW macromolecules and bioparticles.<sup>12</sup>

The present invention provides new dense mineral oxide solid supports or microbeads which exhibit high density, low porosity, high external surface area and high binding capacity. The small dense mineral oxide solid supports or microbeads of the present invention may be used in various solid phase adsorption and chromatography methods including packed bed and fluidized bed methods, and are particularly useful in fluidized bed devices and allow higher linear velocities to be used in such fluidized bed devices. These solid supports or microbeads are particularly suited for separating or isolating large biological molecules, such as bioparticles and high molecule weight macromolecules, especially in fluidized bed or expanded bed methods.<sup>13</sup>

The dense mineral oxide solid supports according to the invention comprise:

(a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and

---

<sup>12</sup> Specification at page 5, lines 7-19.

<sup>13</sup> Specification at page 5, lines 21-30.

(b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support.<sup>14</sup>

In a preferred embodiment, the dense mineral oxide solid supports have a density of 2.1 to 11, and a particle size of 10  $\mu\text{m}$  to 100  $\mu\text{m}$ .<sup>15</sup> The mineral oxide matrix may comprise, *inter alia*, titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof. The interactive polymer network may comprise, *inter alia*, a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.<sup>16</sup>

## ***VI. ISSUES***

The single issue on appeal is whether claims 1-22 and 59-63 would have been obvious over Girod *et al.* (U.S. 5,445,732) alone or combined with Davis, Jr. *et al.* (U.S. 4,203,772).

## ***VII. GROUPS OF CLAIMS***

For purposes of the present appeal, the claims do not all stand or fall together. The following groups of claims will be argued separately:

- |         |   |
|---------|---|
| Group 1 | Claims 1, 9, 10, and 13-22  |
| Group 2 | Claims 11 and 12 (a subset of the claims that recite polysaccharide polymer in the pores) |
| Group 3 | Claims 7, 8 and 60 (a subset of the claims that recite a specified range for pore volume) |
| Group 4 | Claims 2, 3, 6 and 59-63 (a subset of the claims that recite a density for the particles) |

---

<sup>14</sup> See claim 1.

<sup>15</sup> See claim 6.

<sup>16</sup> See claims 9 and 10, respectively.

Thus, the arguments in Section IX.A of the brief apply equally to all of the claims on appeal. The arguments in Sections IX.C.1, IX.C.2, and IX.C.3 of the brief apply to the features that are recited more particularly in Claim Groups 2, 3, and 4, respectively. Accordingly, the arguments advanced in the latter sections additionally support the patentability of claims in Groups 2, 3, and 4, respectively. Even if the arguments that pertain to all appealed claims are not deemed persuasive, the Board is obliged to consider separately the patentability of the claims in each of Groups 2, 3, and 4, in light of the additional arguments presented.

### ***VIII. SUMMARY OF THE ARGUMENT***

Giro *et al.* (U.S. 5,445,732) would not have suggested a dense mineral oxide solid supports comprising a mineral oxide matrix a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support. Both the patent itself and a declaration from one its inventors clarify that the purpose of invention described in Giro ('732) was to make the pore volume as large as possible so that molecules would diffuse inside the pores of the matrix. This is consistent with calculated pore volumes of 40 to 60% for Giro ('732). This approach contrasts sharply with that described in the application, which strives to foreclose access to the pore volume by the molecules in the source solution, using a small pore volume so that a crosslinked polymer can be stably rooted in the matrix and so that interactions should occur on the surface of the bead.

The addition of Davis, Jr. to Giro ('732) does not gainsay this conclusion. While Davis, Jr. discloses mineral oxide matrices having porosities less than 30%, it would not have been obvious to use the matrix material of Davis, Jr. in place of the matrix disclosed in Giro. Such a substitution would be contrary to Giro's purpose of providing beads in which biomolecules penetrate "an open, flexible. . . three-dimensional polymer network" that is contained in the pores. If a proposed modification renders a prior art invention unsatisfactory for its intended purpose, then there is no suggestion or motivation to make

the proposed modification. MPEP §2143.01 and *In re Gordon*, 221 USPQ 1125 (Fed. Cir. 1984).

## ***IX. ARGUMENT***

A. Claims 1-22 and 59-63 would not have been obvious based on Girot *et al.* (U.S. 5,445,732)

Claims 1-22 and 59-63 are rejected under Section 103(a) based on Girot *et al.* The examiner states that:

Girot ('732) discloses dense mineral oxide supports comprising a mineral oxide matrix having an external surface and pores; and an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix. Girot ('732) does not explicitly disclose the porosity being 'less than 30% of the total volume of the mineral oxide matrix' as claimed. However, Girot ('732) recognizes that a broad range of porosities are suitable for the invention [citing col. 8, lines 40-45; col. 15, line 45; and col. 36, lines 12-15]

A dense mineral oxide solid support comprising a mineral oxide matrix having an external surface and pores, in which the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix would not have been suggested by Girot ('732). Girot ('732) discloses, in column 8, that:

Typically, the present invention utilizes base matrices having the following characteristics: an initial average particle size ranging from about 5 to about 1000 microns; an initial porous volume ranging from about 0.2 to about 2 cm<sup>3</sup>/gram; an initial surface area ranging from about 1 to about 800 m<sup>2</sup>/gram; and an initial pore size ranging from about 50 to about 6000 angstroms. Preferably, the base matrix is characterized by: an initial average particle size ranging from about 10 to about 300 microns, although passivated supports having narrow particle size ranges, such as about 15-20, about 15-25, about 30-45, about 50-60, about 80-100, and about 100-300 microns, are most preferred. Preferred ranges for other characteristics include an initial porous volume ranging from about 0.8 to about 1.2 cm<sup>3</sup>/gram; an initial surface area ranging from about 10 to about 400

m<sup>2</sup>/gram; and an initial pore size ranging from about 1000 to about 3000 angstroms. The density of the porous solid matrix obviously varies with its chemical nature, being higher for mineral oxide (*e.g.*, silica) substrates and lower for polymeric ones (*e.g.*, polystyrene).

The use of pore volumes ranging from 0.8 to 1.2 cm<sup>3</sup>/gr of solid material marks a fundamental difference between Girot ('732) and the present invention. Girot ('732) relates primarily to porous silica, for which a porous volume of 0.8 to 1.2 cm<sup>3</sup>/gr equates to a pore volume of between 0.4 to 0.6 ml per ml of solid material or between 40 and 60% (porous silica has a volume/weight ratio of 2, *i.e.*, 2 ml or 2 cm<sup>3</sup> of beads yield about 1 gram of material).

This is consistent with attestations in a declaration of Dr. Boschetti that was submitted during prosecution before the examiner. Dr. Boschetti is a co-inventor of Girot ('732) and thus eminently qualified to inform as to its content. Dr. Boschetti attests that the purpose of invention described in Girot ('732) was to have a hydrogel fill the pores entirely, while allowing even macromolecules still to diffuse inside. Therefore, the pore volume is made as large as possible, to maximize the binding capacity of the final material. The pore volume of 40 to 60% calculated above is in line with this purpose. Viewed in its proper context, which takes into account the purposed of the invention in Girot ('732), the disclosure of Girot ('732) would not have led one of ordinary skill to decrease porosity, or pore volume, below 30%. This approach contrasts sharply with that described in the application. The latter strives to foreclose access to the pore volume by the molecules in the source solution, and it uses a small pore volume in order that a crosslinked polymer can be stably rooted in the matrix and that interactions should occur on the surface of the bead.

This also is supported by an explanation contained in Girot ('732) that relate to the underlying mechanism of the invention. Thus, at column 19 it is disclosed that:

It is believed, without wishing to be limited by theory, that a highly open, flexible lattice structure comprised primarily of polymeric chains of repeating main monomer units is formed within the pores of the porous solid matrix. Very significantly, it is believed that the areas of the porous support available for desirable reversible interaction with



biological molecules are not confined to the regions immediately adjacent to the surface of the pore as is the case when thin, substantially two-dimensional coatings are applied to porous surfaces in the manner of Steuck (U.S. Pat. No. 4,618,533) and Varady *et al.* (U.S. Pat. No. 5,030,352) as discussed in Section 2.2 above. Rather, it is believed that the polymeric network of the present invention extends outwardly into the pore volume itself in the manner of a three-dimensional lattice, as opposed to a two-dimensional coating limited strictly to the pore wall surface area. A schematic diagram of such a structure, as it is thought to exist, is illustrated in FIG. 5, where a biological molecule of interest (depicted as a spherical object) is also shown interacting with the lattice. Furthermore, the presence of porogens (pore-inducers) in the passivation mixture is believed to promote creation of this open three-dimensional polymer network.

It is thought that perhaps the open, flexible nature of the three-dimensional polymer network allows biological molecules *to rapidly penetrate the polymer lattice and thereby efficiently interact with sorptive groups in the polymer network of the passivated porous support* even at high solution flowrates. The *rapid and efficient mass transfer of biomolecules into and through this network* avoids the decrease in useful or dynamic sorption capacity and resolution that are typical of conventional chromatographic media. With these conventional media, diffusion in the pores of the support and/or materials coated thereupon or within them leads to poor mass transfer rates and limits the efficiency of the chromatographic process.

(Emphasis added.)

This approach, in which biomolecules enter the pores of a mineral oxide bead, contrasts sharply with the present invention, in which pore volume is minimized in order to foreclose access to the pore volume by molecules in the solution. The low porosity as presently claimed allows a crosslinked polymer to be stably rooted in the matrix, “so that subsequent interaction with macromolecules occurs on the external surface area of the support.” This is clearly contrary to both the purpose of Girot (‘732) and to the physical structure taught by Girot (‘732).

The examiner argues that the skilled artisan would have been motivated to minimize porosity in light of Girot’s teaching that “it is generally desirable to have as great

a density difference as possible between the solid support particles and the fluidizing medium.” This teaching in Girot (‘732) must be balanced, however, against the primary purpose of Girot (‘732), which was to provide a bead in which even macromolecules can enter the pores. When designing beads for fluidized bed chromatography, the skilled artisan might have been motivated to select values at the lower end of the stated pore volume range in Girot (‘732), in order to maximize the density difference. But the person of ordinary skill would *not* have been motivated to extend the range lower than the endpoint of the disclosed range. Such an extension, beyond the range disclosed in Girot (‘732), would directly contravene the ability to have biomolecules diffuse into the pores. A porosity of less than 30% as presently claimed is therefore not a “workable” range when considered in light of the purpose of Girot (‘732), and one of ordinary skill in the art would not have been motivated to use a pore volume less than the endpoint of Girot’s range.

In an Advisory Action dated July 2, 2003, the examiner states that “the instant claims in no way exclude the possibility of macromolecules entering pores.” The instant claims do recite, however, “an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, *so that subsequent interaction with macromolecules occurs on the external surface area of the support*” – claims 1 and 6, emphasis added. This is in distinct contrast to the mechanism proposed in Girot (‘732), in which molecules diffuse into a polymeric lattice contained within the pores, as quoted above. Subsequent interaction on the external surface area of the support is not suggested in Girot (‘732).

The examiner comments in the Advisory Action that “clearly the size of a given pore at a surface entrance (if there is one) determines if a molecule can enter, not the percent porosity. . . a hollow sphere could have >99% porosity and not let anything in.” Certainly, one can invent hypothetical situations to demonstrate that porosity is not equivalent to openness. However, in the real world of mineral oxide beads a relationship between porosity and the ability of molecules to penetrate is recognized. For example, Girot (‘732) notes that “the present invention relates to the passivation of non-passivated porous solid matrices while *maximizing the openness (e.g., gel porosity and pore size)* of the resulting passivated porous support” (column 9, lines 7-8; emphasis added). The

hypothetical situations proposed in the Advisory Action would not be the basis on which a skilled artisan would interpret the teachings of the prior art.

Finally, the examiner's comments in the Advisory Action that "many claims of Girot ('732) are not limited to any particular porosity" and that "claim 13 of Girot ('732) explicitly recites *excluding* molecules as small as 500 daltons" (emphasis in original) do not belie the clear teaching that interaction in Girot ('732) occurs within the polymeric lattice contained within the pores. The fact that molecules of a certain size are excluded in Girot's claims in no way implies that the molecules of interest, *i.e.*, those not excluded, are interacting on the external surface area of the support as presently claimed. Indeed, the very concept of size exclusion supports the conclusion that Girot's mechanism is one in which molecules diffuse into the beads, as clearly proposed by Girot. This understanding of Girot's mechanism is supported by attestations of one of the inventors of Girot ('732). In light of this, the interpretation by the examiner of Girot's teaching clearly is unsupportable.

B. The addition of Davis, Jr. (U.S. 4,203,772) to Girot ('732) would not have suggested the invention as presently recited in claims 1-22 and 59-63

In the alternative, claims 1-22 and 59-63 stand rejected under Section 103(a) based on Girot ('732) in view of Davis *et al.* (U.S. 4,203,772). The examiner relies on Davis, Jr. *et al.* as explicitly disclosing mineral oxide matrices having porosities less than 30%, and argues that it would have been obvious to use the matrix material of Davis, Jr. *et al.* in place of the matrix disclosed in Girot. Once again, this would be contrary to Girot's purpose of providing beads in which biomolecules penetrate "an open, flexible. . . three-dimensional polymer network" that is contained in the pores. As noted in detail above, interaction in Girot is not on the surface as presently claimed. To modify Girot by using the mineral oxide matrix of Davis would be to render Girot unsuitable for its intended purpose. If a proposed modification renders a prior art invention unsatisfactory for its intended purpose, then perforce there would have been no suggestion or motivation to make the proposed modification. MPEP §2143.01 and *In re Gordon*, 221 USPQ 1125 (Fed. Cir. 1984). Accordingly, the modification proposed by the addition of Davis, Jr. to Girot is improper and cannot be sustained.

C. While no *prima facie* case of obviousness exists for any of the claims, additional bases for patentability exist for subgroups of the appealed claims

1. Claims 11 and 12 recite the pores contain a polysaccharide, a feature not suggested by the art of record

Claim 11 recites the soluble organic polymer is a polysaccharide or a mixture of polysaccharides, while claim 12 more particularly recites that the polysaccharide is selected from the group consisting of agarose, dextran, cellulose, chitosan, a glucosaminoglycan, and derivatives thereof. These polymers are quite different than those in Girot ('732), and would not have been suggested by the cited art of record.

2. Claims 7, 8 and 60 recite a ranges for pore volume that would not have been suggested by the art of record

Claims 7 and 60 recite a pore volume is 5% to 25% of the total volume of the mineral oxide matrix, while claim 8 recites a pore volume of 5% to 15%. These smaller pore volumes deviate even further from Girot's teaching of a maximized pore volume than does the recitation in the other claims of pore volumes less than 30%. Accordingly, the case in favor of patentability is even stronger as to these claims.

3. Claims 2, 3, 6 and 60-63 recite a density for the particles that would not have been suggested by the art of record

Claim 2 recites that the dense mineral oxide solid supports have a density in the range of about 1.7 to 11, claim 3 recites a density in the range of about 2.1 to about 10, and claims 6 and 60-63 recite a density of 2.1 to 11. Girot ('732) merely indicates that "density of the porous solid matrix obviously varies with its chemical nature" (column 8, lines 59-60), and provides no guidance on the issue of density. Accordingly, there clearly is no *prima facie* case of obviousness with respect to claims 2, 3, 6 and 60-63.

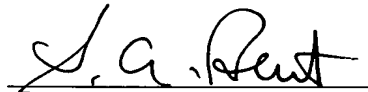
**X. CONCLUSION**

For these reasons, the Board is respectfully requested to reverse the examiner and remand this application for issuance.

Respectfully submitted,

4 November 2003

Date



Stephen A. Bent  
Attorney for Applicant  
Reg. No. 29,768

**FOLEY & LARDNER**

3000 K Street, N.W., Suite 500  
Washington, D.C. 20007-5109  
Telephone: (202) 672-5492  
Facsimile: (202) 672-5399

**APPENDIX: APPEALED CLAIMS**

What is claimed is:

1. (Twice Amended) Dense mineral oxide solid supports comprising:
  - (a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and
  - (b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support.
2. The dense mineral oxide solid supports of Claim 1, having a density in the range of about 1.7 to 11.
3. The dense mineral oxide solid supports of Claim 2, wherein the density is in the range of about 2.1 to about 10.
4. The dense mineral oxide solid supports of Claim 1, wherein said dense mineral oxide solid supports have a particle size in the range of about 5  $\mu\text{m}$  to about 500  $\mu\text{m}$ .
5. The dense mineral oxide solid supports of Claim 4, wherein the particle size is in the range of about 10  $\mu\text{m}$  to about 100  $\mu\text{m}$ .
6. (Twice Amended) Dense mineral oxide solid supports comprising:
  - (a) a mineral oxide matrix having an external surface and pores, wherein the pores have a pore volume which is less than 30% of the total volume of the mineral oxide matrix, and
  - (b) an interactive polymer network which fills the pores and is coated on the surface of the mineral oxide matrix, so that subsequent interaction with macromolecules occurs on the external surface area of the support,

wherein said dense mineral oxide solid supports have a density of 2.1 to 11, and a particle size of 10  $\mu\text{m}$  to 100  $\mu\text{m}$ .

7. The dense mineral oxide solid supports of claim 1, wherein the pore volume is 5% to 25% of the total volume of the mineral oxide matrix.

8. The dense mineral oxide solid supports of Claim 7, wherein the pore volume is 5% to 15%.

9. The dense mineral oxide solid supports of claim 1, wherein the mineral oxide matrix is comprised of titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof.

10. The dense mineral oxide solid supports of claim 1, wherein the interactive polymer network comprises a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.

11. The dense mineral oxide solid supports of Claim 10, wherein the soluble organic polymer is a polysaccharide or a mixture of polysaccharides.

12. The dense mineral oxide solid supports of Claim 11, wherein the polysaccharide is selected from the group consisting of agarose, dextran, cellulose, chitosan, a glucosaminoglycan, and derivatives thereof.

13. The dense mineral oxide solid supports of Claim 10, wherein the soluble organic polymer is a linear soluble organic polymer selected from the group consisting of polyvinyl alcohol, a polyethyleneimine, a polyvinylamine, polyvinylpyrrolidone, a polyethyleneglycol, a polyaminoacid, a polynucleic acid, and derivatives thereof.

14. The dense mineral oxide solid supports of claim 1, wherein the interactive polymer network comprises monomers, bifunctional monomers, or mixtures thereof copolymerized in place with the mineral oxide matrix.

15. The dense mineral oxide solid supports of Claim 14, wherein the monomers are selected from the group consisting of:

(a) aliphatic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds;

(b) aromatic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds;

(c) heterocyclic ionic, non-ionic, and reactive derivatives of acrylic, methacrylic, vinylic, and allylic compounds; and

(d) mixtures of any of the monomers in (a), (b) or (c).

16. The dense mineral oxide solid supports of Claim 15, wherein (a) is acrylamide, dimethylacrylamide, trisacryl, acrylic acid, acryloylglycine, diethylaminoethyl methacrylamide, vinylpyrrolidone, vinylsulfonic acid, allylamine, allylglycidylether, or derivatives thereof.

17. The dense mineral oxide solid supports of Claim 15, wherein (b) is vinyltoluene, phenylpropylacrylamide, trimethylaminophenylbutylmethacrylate, tritylacrylamid; or derivatives thereof.

18. The dense mineral oxide solid supports of Claim 15, wherein (c) is vinylimidazole, vinylpyrrolidone, acryloylmorpholine, or derivatives thereof.

19. The dense mineral oxide solid supports of Claim 14, wherein the bifunctional monomers are selected from the group consisting of:

(a) bisacrylamides;

(b) bis-methacrylamides;

(c) bis-acrylates;

(d) ethyleneglycol-methacrylates; and

(e) diallyltartrdiamide.

20. The dense mineral oxide solid supports of Claim 19, wherein (a) is N,N'-methylene-bis-acrylamide, N,N'-ethylene-bis-acrylamide, N,N'-hexamethylene-bis-acrylamide, or glyoxal-bis-acrylamide.



21. The dense mineral oxide solid supports of Claim 19, wherein (b) is N,N'-methylene-bis-methacrylamide, N,N'-ethylene-bis-methacrylamide, or N,N'-hexamethylene-bis-methacrylamide.

22. The dense mineral oxide solid supports of Claim 19, wherein (c) is ethyleneglycoldiacrylate, or ethyleneglycoldimethacrylate.

59. The dense mineral oxide solid supports of claim 2, wherein said dense mineral oxide solid supports have a particle size in the range of about 5  $\mu\text{m}$  to about 500  $\mu\text{m}$ .

60. The dense mineral oxide solid supports of claim 6, wherein the pore volume is 5% to 25% of the total volume of the mineral oxide matrix.

61. The dense mineral oxide solid supports of claim 6, wherein the mineral oxide matrix is comprised of titania, zirconia, yttria, ceria, hafnia, tantalia, or mixtures thereof.

62. The dense mineral oxide solid supports of claim 6, wherein the interactive polymer network comprises a soluble organic polymer or a mixture of soluble organic polymers crosslinked in place with the mineral oxide matrix.

63. The dense mineral oxide solid supports of claim 6, wherein the interactive polymer network comprises monomers, bifunctional monomers, or mixtures thereof copolymerized in place with the mineral oxide matrix.